

=> s pseudomonas (w) fluorescens and alginate and mutant
L10 38 PSEUDOMONAS (W) FLUORESCENS AND ALGINATE AND MUTANT

=> s pseudomonas (w) fluorescens and alginate and mutant (w) strain
L11 2 PSEUDOMONAS (W) FLUORESCENS AND ALGINATE AND MUTANT (W) STRAIN

=> d ibib abs l10 1-38

L10 ANSWER 1 OF 38 MEDLINE on STN
ACCESSION NUMBER: 2003447486 MEDLINE
DOCUMENT NUMBER: PubMed ID: 14507360
TITLE: Biofilm formation at the air-liquid interface by the
Pseudomonas fluorescens SBW25 wrinkly
spreader requires an acetylated form of cellulose.
AUTHOR: Spiers Andrew J; Bohannon John; Gehrig Stefanie M; Rainey
Paul B
CORPORATE SOURCE: Department of Plant Sciences, University of Oxford, South
Parks Road, Oxford OX1 3RB, UK..
andrew.spiers@plants.ox.ac.uk
SOURCE: Molecular microbiology, (2003 Oct) Vol. 50, No. 1, pp.
15-27.
Journal code: 8712028. ISSN: 0950-382X.
PUB. COUNTRY: England: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200312
ENTRY DATE: Entered STN: 26 Sep 2003
Last Updated on STN: 18 Dec 2003
Entered Medline: 10 Dec 2003

AB The wrinkly spreader (WS) genotype of Pseudomonas fluorescens SBW25 colonizes the air-liquid interface of spatially structured microcosms resulting in formation of a thick biofilm. Its ability to colonize this niche is largely due to overproduction of a cellulosic polymer, the product of the wss operon. Chemical analysis of the biofilm matrix shows that the cellulosic polymer is partially acetylated cellulose, which is consistent with predictions of gene function based on in silico analysis of wss. Both polar and non-polar mutations in the sixth gene of the wss operon (wssF) or adjacent downstream genes (wssGHIJ) generated mutants that overproduce non-acetylated cellulose, thus implicating WssFGHIJ in acetylation of cellulose. WssGHI are homologues of AlgFIJ from P. aeruginosa, which together are necessary and sufficient to acetylate alginate polymer. WssF belongs to a newly established Pfam family and is predicted to provide acyl groups to WssGHI. The role of WssJ is unclear, but its similarity to MinD-like proteins suggests a role in polar localization of the acetylation complex. Fluorescent microscopy of Calcofluor-stained biofilms revealed a matrix structure composed of networks of cellulose fibres, sheets and clumped material. Quantitative analyses of biofilm structure showed that acetylation of cellulose is important for effective colonization of the air-liquid interface: mutants identical to WS, but defective in enzymes required for acetylation produced biofilms with altered physical properties. In addition, mutants producing non-acetylated cellulose were unable to spread rapidly across solid surfaces. Inclusion in these assays of a WS mutant with a defect in the GGDEF regulator (WspR) confirmed the requirement for this protein in expression of both acetylated cellulose polymer and bacterial attachment. These results suggest a model in which WspR regulation of cellulose expression and attachment plays a role in the co-ordination of surface colonization.

L10 ANSWER 2 OF 38 MEDLINE on STN
ACCESSION NUMBER: 2003273771 MEDLINE

DOCUMENT NUMBER: PubMed ID: 12775688
 TITLE: The *Pseudomonas fluorescens* AlgG protein, but not its mannuronan C-5-epimerase activity, is needed for alginate polymer formation.
 AUTHOR: Gimmestad Martin; Sletta Havard; Ertesvag Helga; Bakkevig Karianne; Jain Sumita; Suh Sang-jin; Skjak-Braek Gudmund; Ellingsen Trond E; Ohman Dennis E; Valla Svein
 CORPORATE SOURCE: Department of Biotechnology, Norwegian University of Science and Technology, Trondheim, Norway.
 CONTRACT NUMBER: AI-19146 (NIAID)
 SOURCE: Journal of bacteriology, (2003 Jun) Vol. 185, No. 12, pp. 3515-23.
 Journal code: 2985120R. ISSN: 0021-9193.
 PUB. COUNTRY: United States
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
 LANGUAGE: English
 FILE SEGMENT: Priority Journals
 OTHER SOURCE: GENBANK-AF527790
 ENTRY MONTH: 200307
 ENTRY DATE: Entered STN: 13 Jun 2003
 Last Updated on STN: 8 Jul 2003
 Entered Medline: 7 Jul 2003

AB Bacterial alginates are produced as 1-4-linked beta-D-mannuronan, followed by epimerization of some of the mannuronic acid residues to alpha-L-guluronic acid. Here we report the isolation of four different epimerization-defective point mutants of the periplasmic *Pseudomonas fluorescens* mannuronan C-5-epimerase AlgG. All mutations affected amino acids conserved among AlgG-epimerases and were clustered in a part of the enzyme also sharing some sequence similarity to a group of secreted epimerases previously reported in *Azotobacter vinelandii*. An algG-deletion mutant was constructed and found to produce predominantly a dimer containing a 4-deoxy-L-erythro-hex-4-enepyransyluronate residue at the nonreducing end and a mannuronic acid residue at the reducing end. The production of this dimer is the result of the activity of an alginate lyase, AlgL, whose in vivo activity is much more limited in the presence of AlgG. A strain expressing both an epimerase-defective (point mutation) and a wild-type epimerase was constructed and shown to produce two types of alginate molecules: one class being pure mannuronan and the other having the wild-type content of guluronic acid residues. This formation of two distinct classes of polymers in a genetically pure cell line can be explained by assuming that AlgG is part of a periplasmic protein complex.

L10 ANSWER 3 OF 38 MEDLINE on STN
 ACCESSION NUMBER: 2001654609 MEDLINE
 DOCUMENT NUMBER: PubMed ID: 11707327
 TITLE: Characterization of algG encoding C5-epimerase in the alginate biosynthetic gene cluster of *Pseudomonas fluorescens*.
 AUTHOR: Morea A; Mathee K; Franklin M J; Giacomini A; O'Regan M; Ohman D E
 CORPORATE SOURCE: CRIBI, Biotechnology Centre, University of Padova, Viale G. Colombo 3, 35121 Padova, Italy.
 CONTRACT NUMBER: AI-19146 (NIAID)
 SOURCE: Gene, (2001 Oct 31) Vol. 278, No. 1-2, pp. 107-14.
 Journal code: 7706761. ISSN: 0378-1119.
 PUB. COUNTRY: Netherlands
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
 LANGUAGE: English
 FILE SEGMENT: Priority Journals
 ENTRY MONTH: 200201
 ENTRY DATE: Entered STN: 15 Nov 2001
 Last Updated on STN: 25 Jan 2002
 Entered Medline: 10 Jan 2002

AB The organization of the alginate gene cluster in *Pseudomonas fluorescens* was characterized. A bank of genomic DNA from *P. fluorescens* was mobilized to a strain of *Pseudomonas aeruginosa* with a transposon insertion (*algJ::Tn501*) in the alginate biosynthetic operon that rendered it non-mucoid. Phenotypic complementation in this heterologous host was observed, and a complementing clone containing 32 kb of *P. fluorescens* DNA was obtained. Southern hybridization studies showed that genes involved in alginate biosynthesis (e.g. *algD*, *algG*, and *algA*) were approximately in the same order and position as in *P. aeruginosa*. When the clone was mobilized to a *P. aeruginosa* *algG* mutant that produced alginate as polymannuronate due to its C5-epimerase defect, complementation was observed and the alginate from the recombinant strain contained L-guluronate as determined by proton nuclear magnetic resonance spectroscopy. A sequence analysis of the *P. fluorescens* DNA containing *algG* revealed sequences similar to *P. aeruginosa* *algG* that were also flanked by *algE*- and *algX*-like sequences. The predicted *AlgG* amino acid sequence of *P. fluorescens* was 67% identical (80% similar) to *P. aeruginosa* *AlgG* and 60% identical (76% similar) to *Azotobacter vinelandii* *AlgG*. As in *P. aeruginosa*, *AlgG* from *P. fluorescens* appeared to have a signal sequence that would localize it to the periplasm where *AlgG* presumably acts as a C5-epimerase at the polymer level. Non-polar *algG* knockout mutants of *P. fluorescens* were defective in alginate production, suggesting a potential role for this protein in polymer formation.

L10 ANSWER 4 OF 38 MEDLINE on STN
 ACCESSION NUMBER: 82144389 MEDLINE
 DOCUMENT NUMBER: PubMed ID: 6801192
 TITLE: Isolation of alginate-producing mutants of *Pseudomonas fluorescens*, *Pseudomonas putida* and *Pseudomonas mendocina*.
 AUTHOR: Govan J R; Fyfe J A; Jarman T R
 SOURCE: Journal of general microbiology, (1981 Jul) Vol. 125, No. 1, pp. 217-20.
 Journal code: 0375371. ISSN: 0022-1287.
 PUB. COUNTRY: ENGLAND: United Kingdom
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
 LANGUAGE: English
 FILE SEGMENT: Priority Journals
 ENTRY MONTH: 198205
 ENTRY DATE: Entered STN: 17 Mar 1990
 Last Updated on STN: 3 Feb 1997
 Entered Medline: 21 May 1982

AB Spontaneous alginate-producing (*muc*) variants were isolated from strains of *Pseudomonas fluorescens*, *P. putida* and *P. mendocina* at a frequency of 1 in 10(8) by selecting for carbenicillin resistance. The infrared spectrum of the bacterial exopolysaccharide was typical of an acetylated alginate similar to that previously described in *Azotobacter vinelandii* and in mucoid variants of *P. aeruginosa*. Mucoid variants were not isolated from *P. stutzeri*, *P. pseudoalcaligenes*, *P. testosteroni*, *P. diminuta*, *P. acidovorans*, *P. cepacia* or *P. maltophilia*.

L10 ANSWER 5 OF 38 AGRICOLA Compiled and distributed by the National Agricultural Library of the Department of Agriculture of the United States of America. It contains copyrighted materials. All rights reserved.
 (2006) on STN

ACCESSION NUMBER: 93:76265 AGRICOLA
 DOCUMENT NUMBER: IND93050364
 TITLE: Influence of *Pseudomonas fluorescens* on hyphal growth and biocontrol activity of *Trichoderma harizanum* in the spermosphere and rhizosphere of pea.

AUTHOR(S): Dandurand, L.M.; Knudsen, G.R.
AVAILABILITY: DNAL (464.8 P56)
SOURCE: Phytopathology, Mar 1993. Vol. 83, No. 3. p. 265-270
Publisher: St. Paul, Minn. : American
Phytopathological Society.
CODEN: PHYTAJ; ISSN: 0031-949X
NOTE: Includes references.

DOCUMENT TYPE: Article
FILE SEGMENT: U.S. Imprints not USDA, Experiment or Extension
LANGUAGE: English

AB Trichoderma harzianum isolate ThzID1 was grown in liquid culture, was formulated with alginate and polyethylene glycol 8000; and was milled into fine granules (average diameter 500 micrometers). Granules contained chlamydospores, conidia, and hyphal fragments. Viability of the encapsulated fungus remained high for at least 6 mo when stored at 5 C (i.e., > 90% of the granules produced hyphal growth when incubated on agar); viability was reduced significantly when granules were stored at 22 C. Application of the granular formulation of T. harzianum to pea seeds reduced root rot by Aphanomyces euteiches f. sp. pisi in growth-chamber experiments and also increased plant top weights compared to noncoated seeds. Seed treatment with slurries of Pseudomonas fluorescens strain 2-79RN(10), which produces a phenazine antibiotic, also reduced Aphanomyces root rot but to a lesser extent than did T. harzianum ThzID1. Disease suppression was not significantly different when seeds were treated with a combination of T. harzianum and 2-79RN(10) compared to treatment with T. harzianum alone. Root rot was not reduced by the mutant P. fluorescens strain 2-79-B46, which lacks phenazine. Treatment with T. harzianum plus 2-79-B46 resulted in the same level of disease control achieved by T. harzianum alone. These results suggest that the biocontrol mechanism of P. fluorescens 2-79RN(10) neither inhibited nor enhanced the biocontrol activity of T. harzianum ThzID1. In other experiments, density of T. harzianum hyphae originating from coated pea seeds in soil was not affected by the addition of 2-79RN(10), but when 2-79-B46 was added, density was greater after 5 days. The colony radius of T. harzianum was initially enhanced (at 3 days) by the addition of either strain, but the effect diminished by day 5. The same treatments were then applied to peas and to glass beads of equivalent size, and similar effects of the added bacterial strains were observed on both substrates, suggesting that the growth enhancement of T. harzianum in the presence of bacteria was not the direct result of stimulation of seed exudation by the bacteria. Our results provide a potentially improved formulation methodology for coating seeds with biocontrol organisms and methods for evaluating the compatibility of fungal and bacterial biocontrol agents applied to seeds.

L10 ANSWER 6 OF 38 AGRICOLA Compiled and distributed by the National Agricultural Library of the Department of Agriculture of the United States of America. It contains copyrighted materials. All rights reserved.
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ACCESSION NUMBER: 91:81096 AGRICOLA
DOCUMENT NUMBER: IND91045062
TITLE: Influence of an antagonistic strain of Pseudomonas fluorescens on growth and ability of Trichoderma harzianum to colonize sclerotia of Sclerotinia sclerotiorum in soil.
AUTHOR(S): Bin, L.; Knudsen, G.R.; Eschen, D.J.
CORPORATE SOURCE: University of Idaho, Moscow
AVAILABILITY: DNAL (464.8 P56)
SOURCE: Phytopathology, Sept 1991. Vol. 81, No. 9. p. 994-1000
Publisher: St. Paul, Minn. : American
Phytopathological Society.
CODEN: PHYTAJ; ISSN: 0031-949X
NOTE: Includes references.
DOCUMENT TYPE: Article

FILE SEGMENT: U.S. Imprints not USDA, Experiment or Extension
LANGUAGE: English

AB *Pseudomonas fluorescens* strain 2-79RN10 (nalidixic acid and rifampicin-resistant mutant of wild type strain 2-79) was used to study potential effects of bacterial antagonism in soil on growth and biocontrol efficacy of the biocontrol fungus *Trichoderma harzianum* isolate THzID1, which was formulated into alginate pellets. In steamed soil (25 C, -100 or -500 kPa matric potential), strain 2-79RN10 maintained its initial high populations (approximately 3×10^4 or 3×10^7 cfu/g of soil) over a 14-day period, and significantly reduced hyphal radius, hyphal density, and recoverable numbers of propagules of ThzID1. In raw soil under similar environmental conditions (22-25 C, -10 to -1,000 kPa), populations of 2-79RN10 decreased by approximately four log₁₀ units over a 3-wk period, and did not affect the ability of *Trichoderma* spp. to colonize sclerotia of *Sclerotinia sclerotiorum*. In two years of field experiments using raw or steamed soil in microplots, populations of 2-79RN10 decreased gradually after 1-2 wk and did not reduce the ability of *Trichoderma* spp. to colonize sclerotia of *S. sclerotiorum*. Colonization of sclerotia by *Trichoderma* spp. after 9 wk was significantly higher in steamed soil (mean = 65%) than in raw soil (mean = 30%) when THzID1 was added, suggesting possible inhibition of THzID1 by indigenous soil microbes, or utilization by ThzID1 of nutrients released by steaming of soil. In treatments where ThzID1 was not added, low levels of colonization of sclerotia were observed, apparently due to indigenous *Trichoderma* spp., and these levels were higher in raw soil (mean = 18%) than in steamed soil (mean = 5%). These results suggest that under certain restrictive conditions, high population levels of antagonistic bacteria in bulk soil suppressed a fungal biocontrol agent, but that this suppressive effect was reduced or eliminated when a high bacterial population was not present.

L10 ANSWER 7 OF 38 BIOSIS COPYRIGHT (c) 2006 The Thomson Corporation on STN
ACCESSION NUMBER: 2003:510388 BIOSIS
DOCUMENT NUMBER: PREV200300497223
TITLE: Biofilm formation at the air-liquid interface by the *Pseudomonas fluorescens* SBW25 wrinkly spreader requires an acetylated form of cellulose.
AUTHOR(S): Spiers, Andrew J. [Reprint Author]; Bohannon, John; Gehrig, Stefanie M.; Rainey, Paul B.
CORPORATE SOURCE: Department of Plant Sciences, University of Oxford, South Parks Road, Oxford, OX1 3RB, UK
andrew.spiers@plants.ox.ac.uk
SOURCE: Molecular Microbiology, (October 2003) Vol. 50, No. 1, pp. 15-27. print.
ISSN: 0950-382X (ISSN print).
DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 29 Oct 2003
Last Updated on STN: 29 Oct 2003

AB The wrinkly spreader (WS) genotype of *Pseudomonas fluorescens* SBW25 colonizes the air-liquid interface of spatially structured microcosms resulting in formation of a thick biofilm. Its ability to colonize this niche is largely due to overproduction of a cellulosic polymer, the product of the wss operon. Chemical analysis of the biofilm matrix shows that the cellulosic polymer is partially acetylated cellulose, which is consistent with predictions of gene function based on in silico analysis of wss. Both polar and non-polar mutations in the sixth gene of the wss operon (wssF) or adjacent downstream genes (wssGHIJ) generated mutants that overproduce non-acetylated cellulose, thus implicating WssFGHIJ in acetylation of cellulose. WssGHI are homologues of AlgFIJ from *P. aeruginosa*, which together are necessary and sufficient to acetylate alginate polymer. WssF belongs to a newly established Pfam family and is predicted to provide acyl groups to WssGHI. The role of WssJ is unclear, but its

similarity to MinD-like proteins suggests a role in polar localization of the acetylation complex. Fluorescent microscopy of Calcofluor-stained biofilms revealed a matrix structure composed of networks of cellulose fibres, sheets and clumped material. Quantitative analyses of biofilm structure showed that acetylation of cellulose is important for effective colonization of the air-liquid interface: mutants identical to WS, but defective in enzymes required for acetylation produced biofilms with altered physical properties. In addition, mutants producing non-acetylated cellulose were unable to spread rapidly across solid surfaces. Inclusion in these assays of a WS mutant with a defect in the GGDEF regulator (WspR) confirmed the requirement for this protein in expression of both acetylated cellulose polymer and bacterial attachment. These results suggest a model in which WspR regulation of cellulose expression and attachment plays a role in the co-ordination of surface colonization.

L10 ANSWER 8 OF 38 BIOSIS COPYRIGHT (c) 2006 The Thomson Corporation on STN
 ACCESSION NUMBER: 2003:303128 BIOSIS
 DOCUMENT NUMBER: PREV200300303128
 TITLE: The *Pseudomonas fluorescens* AlgG protein, but not its mannuronan C-5-epimerase activity, is needed for alginate polymer formation.
 AUTHOR(S): Gimmestad, Martin; Sletta, Havard; Ertesvag, Helga; Bakkevig, Karianne; Jain, Sumita; Suh, Sang-Jin; Skjak-Braek, Gudmund; Ellingsen, Trond E.; Ohman, Dennis E.; Valla, Svein [Reprint Author]
 CORPORATE SOURCE: Department of Biotechnology, NTNU Norwegian University of Science and Technology, N-7491, Trondheim, Norway svein.valla@biotech.ntnu.no
 SOURCE: Journal of Bacteriology, (June 2003) Vol. 185, No. 12, pp. 3515-3523. print.
 CODEN: JOBAAY. ISSN: 0021-9193.
 DOCUMENT TYPE: Article
 LANGUAGE: English
 ENTRY DATE: Entered STN: 2 Jul 2003
 Last Updated on STN: 2 Jul 2003

AB Bacterial alginates are produced as 1-4-linked beta-D-mannuronan, followed by epimerization of some of the mannuronic acid residues to alpha-L-guluronic acid. Here we report the isolation of four different epimerization-defective point mutants of the periplasmic *Pseudomonas fluorescens* mannuronan C-5-epimerase AlgG. All mutations affected amino acids conserved among AlgG-epimerases and were clustered in a part of the enzyme also sharing some sequence similarity to a group of secreted epimerases previously reported in *Azotobacter vinelandii*. An algG-deletion mutant was constructed and found to produce predominantly a dimer containing a 4-deoxy-L-erythro-hex-4-enepyranosyluronate residue at the nonreducing end and a mannuronic acid residue at the reducing end. The production of this dimer is the result of the activity of an alginate lyase, AlgL, whose in vivo activity is much more limited in the presence of AlgG. A strain expressing both an epimerase-defective (point mutation) and a wild-type epimerase was constructed and shown to produce two types of alginate molecules: one class being pure mannuronan and the other having the wild-type content of guluronic acid residues. This formation of two distinct classes of polymers in a genetically pure cell line can be explained by assuming that AlgG is part of a periplasmic protein complex.

L10 ANSWER 9 OF 38 BIOSIS COPYRIGHT (c) 2006 The Thomson Corporation on STN
 ACCESSION NUMBER: 2002:41402 BIOSIS
 DOCUMENT NUMBER: PREV200200041402
 TITLE: Characterization of algG encoding C5-epimerase in the alginate biosynthetic gene cluster of *Pseudomonas fluorescens*.
 AUTHOR(S): Morea, Antonella; Mathee, Kalai; Franklin, Michael J.;

Giacomini, Alessio; O'Regan, Michael; Ohman, Dennis E.
[Reprint author]
CORPORATE SOURCE: Department of Microbiology and Immunology, Medical College,
Virginia Commonwealth University, 1101 E. Marshall Street,
Virginia Campus, 5-047 Sanger Hall, Richmond, VA,
23298-0678, USA
deohman@hsc.vcu.edu
SOURCE: Gene (Amsterdam), (31 October, 2001) Vol. 278, No. 1-2, pp.
107-114. print.
CODEN: GENED6. ISSN: 0378-1119.
DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 2 Jan 2002
Last Updated on STN: 25 Feb 2002

AB The organization of the alginate gene cluster in
Pseudomonas fluorescens was characterized. A bank of
genomic DNA from *P. fluorescens* was mobilized to a strain of *Pseudomonas*
aeruginosa with a transposon insertion (*algJ::Tn501*) in the
alginate biosynthetic operon that rendered it non-mucoid.
Phenotypic complementation in this heterologous host was observed, and a
complementing clone containing 32 kb of *P. fluorescens* DNA was obtained.
Southern hybridization studies showed that genes involved in
alginate biosynthesis (e.g. *algD*, *algG*, and *algA*) were
approximately in the same order and position as in *P. aeruginosa*. When
the clone was mobilized to a *P. aeruginosa* *algG* mutant that
produced alginate as polymannuronate due to its C5-epimerase
defect, complementation was observed and the alginate from the
recombinant strain contained L-guluronate as determined by proton nuclear
magnetic resonance spectroscopy. A sequence analysis of the *P.*
fluorescens DNA containing *algG* revealed sequences similar to *P.*
aeruginosa *algG* that were also flanked by *algE*- and *algX*-like sequences.
The predicted *AlgG* amino acid sequence of *P. fluorescens* was 67% identical
(80% similar) to *P. aeruginosa* *AlgG* and 60% identical (76% similar) to
Azotobacter vinelandii *AlgG*. As in *P. aeruginosa*, *AlgG* from *P.*
fluorescens appeared to have a signal sequence that would localize it to
the periplasm where *AlgG* presumably acts as a C5-epimerase at the polymer
level. Non-polar *algG* knockout mutants of *P. fluorescens* were
defective in alginate production, suggesting a potential role
for this protein in polymer formation.

L10 ANSWER 10 OF 38 BIOSIS COPYRIGHT (c) 2006 The Thomson Corporation on
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ACCESSION NUMBER: 1993:233118 BIOSIS
DOCUMENT NUMBER: PREV199395124293
TITLE: Influence of *Pseudomonas fluorescens* on
hyphal growth and biocontrol activity of *Trichoderma*
harzianum in the spermosphere and rhizosphere of pea.
AUTHOR(S): Dandurand, L. M.; Knudsen, G. R.
CORPORATE SOURCE: Plant Pathol. Div., Univ. Idaho, Moscow, ID 83843, USA
SOURCE: Phytopathology, (1993) Vol. 83, No. 3, pp. 265-270.
CODEN: PHYTAJ. ISSN: 0031-949X.
DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 7 May 1993
Last Updated on STN: 7 May 1993

AB *Trichoderma harzianum* isolate ThzID1 was grown in liquid culture, was
formulated with alginate and polyethylene glycol 18000, and was
milled into fine granules (average diameter 500 μ -m). Granules contained
chlamydospores, conidia, and hyphal fragments. Viability of the
encapsulated fungus remained high for at least 6 mo when stored at 5 C
(i.e., gt 90% of the granules produced hyphal growth when incubated on
agar); viability was reduced significantly when granules were stored at 22
C. Application of the granular formulation of *T. harzianum* to pea seeds
reduced root rot by *Aphanomyces euteiches* f. sp. *pisii* in growth-chamber

experiments and also increased plant top weights compared to noncoated seeds. Seed treatment with slurries of *Pseudomonas fluorescens* strain 2-79RN-10, which produces a phenazine antibiotic, also reduced *Aphanomyces* root rot but to a lesser extent than did *T. harzianum* ThzID1. Disease suppression was not significantly different when seeds were treated with a combination of *T. harzianum* and 2-79RN-10 compared to treatment with *T. harzianum* alone. Root rot was not reduced by the mutant *P. fluorescens* strain 2-79-B46, which lacks phenazine. Treatment with *T. harzianum* plus 2-79-B46 resulted in the same level of disease control achieved by *T. harzianum* alone. These results suggest that the biocontrol mechanism of *P. fluorescens* 2-79RN-10 neither inhibited nor enhanced the biocontrol activity of *T. harzianum* ThzID1. In other experiments, density of *T. harzianum* hyphae originating from coated pea seeds in soil was not affected by the addition of 2-79RN-10, but when 2-79-B46 was added, density was greater after 5 days. The colony radius of *T. harzianum* was initially enhanced (at 3 days) by the addition of either strain, but the effect diminished by day 5. The same treatments were then applied to peas and to glass beads of equivalent size, and similar effects of the added bacterial strains were observed on both substrates, suggesting that the growth enhancement of *T. harzianum* in the presence of bacteria was not the direct result of stimulation of seed germination by the bacteria. Our results provide a potentially improved formulation methodology for coating seeds with biocontrol organisms and methods for evaluating the compatibility of fungal and bacterial biocontrol agents applied to seeds.

L10 ANSWER 11 OF 38 BIOSIS COPYRIGHT (c) 2006 The Thomson Corporation on STN

ACCESSION NUMBER: 1992:361205 BIOSIS
DOCUMENT NUMBER: PREV199243039355; BR43:39355
TITLE: BIOCONVERSION OF VANILLIN INTO VANILLIC ACID BY
PSEUDOMONAS-FLUORESCENS STRAIN BTP9 CELL
REACTORS AND MUTANTS STUDY.
AUTHOR(S): BARE G [Reprint author]; GERARD J; JACQUES P; DELAUNOIS V;
THONART P
CORPORATE SOURCE: CENTRE WALLON BIOL IND, UNIV LIEGE, FSA GX, SART-TILMAN
B40, 4000 LIEGE, BELG
SOURCE: Applied Biochemistry and Biotechnology, (1992) Vol. 34-35,
pp. 499-514.
Meeting Info.: THIRTEENTH SYMPOSIUM ON BIOTECHNOLOGY FOR
FUELS AND CHEMICALS, COLORADO SPRINGS, COLORADO, USA, MAY
6-10, 1991. APPL BIOCHEM BIOTECHNOL.
CODEN: ABIBDL. ISSN: 0273-2289.
DOCUMENT TYPE: Conference; (Meeting)
FILE SEGMENT: BR
LANGUAGE: ENGLISH
ENTRY DATE: Entered STN: 30 Jul 1992
Last Updated on STN: 30 Jul 1992

L10 ANSWER 12 OF 38 BIOSIS COPYRIGHT (c) 2006 The Thomson Corporation on STN

ACCESSION NUMBER: 1991:482821 BIOSIS
DOCUMENT NUMBER: PREV199192116581; BA92:116581
TITLE: INFLUENCE OF AN ANTAGONISTIC STRAIN OF PSEUDOMONAS
-FLUORESCENS ON GROWTH AND ABILITY OF
TRICHODERMA-HARZIANUM TO COLONIZE SCLEROTIA OF
SCLEROTINIA-SCLEROTIUM IN SOIL.
AUTHOR(S): BIN L [Reprint author]; KNUDSEN G R; ESCHEN D J
CORPORATE SOURCE: PLANT PATHOL DIV, DEP PLANT SOIL AND ENTOMOLOGICAL SCI,
UNIV IDAHO, MOSCOW, IDAHO 83843, USA
SOURCE: Phytopathology, (1991) Vol. 81, No. 9, pp. 994-1000.
CODEN: PHYTAJ. ISSN: 0031-949X.
DOCUMENT TYPE: Article
FILE SEGMENT: BA

LANGUAGE: ENGLISH

ENTRY DATE: Entered STN: 26 Oct 1991

Last Updated on STN: 26 Oct 1991

AB *Pseudomonas fluorescens* strain 2-79N10 (nalidixic acid and rifampicin-resistant mutant of wild type strain 2-79) was used to study potential effects of bacterial antagonism in soil on growth and biocontrol efficacy of the biocontrol fungus *Trichoderma harzianum* isolate ThzID1, which was formulated into alginate pellets. In steamed soil (25 C, -100 or -500 kPa matric potential), strain 2-79N10 maintained its initial high populations (approximately 3×10^4 or 3×10^7 cfu/g of soil) over a 14-day period, and significantly reduced hyphal radius, hyphal density, and recoverable numbers of propagules of ThzID1. In raw soil under similar environmental conditions (22-25 C, -10 to -1,000 kPa), populations of 2-79N10 decreased by approximately four log₁₀ units over a 3-wk period, and did not affect the ability of *Trichoderma* spp. to colonize sclerotia of *Sclerotinia sclerotiorum*. In two years of field experiments using a raw or steamed soil in microplots, populations of 2-79N10 decreased gradually after 1-2 wk and did not reduce the ability of *Trichoderma* spp. to colonize sclerotia *S. sclerotiorum*. Colonization of sclerotia by *Trichoderma* spp. after 9 wk was significantly higher in steamed soil (mean = 65%) than in raw soil (mean = 30%) when ThzID1 was added, suggesting possible inhibition of ThzID1 by indigenous soil microbes, or utilization by ThzID1 of nutrients released by steaming of soil. In treatments where ThzID1 was not added, low levels of colonization of sclerotia were observed, apparently due to indigenous *Trichoderma* spp., and these levels were higher in raw soil (mean = 18%) than in steamed soil (mean = 5%). These results suggest that under certain restrictive conditions, high population levels of antagonistic bacteria in bulk soil suppressed a fungal biocontrol agent, but that this suppressive effect was reduced or eliminated when a high bacterial population was not present.

L10 ANSWER 13 OF 38 BIOSIS COPYRIGHT (c) 2006 The Thomson Corporation on STN

ACCESSION NUMBER: 1984:195488 BIOSIS

DOCUMENT NUMBER: PREV198477028472; BA77:28472

TITLE: SELECTION OF ATTACHMENT MUTANTS DURING THE CONTINUOUS CULTURE OF *PSEUDOMONAS-FLUORESCENS* AND RELATIONSHIP BETWEEN ATTACHMENT ABILITY AND SURFACE COMPOSITION.

AUTHOR(S): PRINGLE J H [Reprint author]; FLETCHER M; ELLWOOD D C

CORPORATE SOURCE: DEP OF MOLECULAR BIOL, UNIV OF EDINBURGH, EDINBURGH EH9 3JR, UK

SOURCE: Journal of General Microbiology, (1983) Vol. 129, No. 8, pp. 2557-2570.

CODEN: JGMIAN. ISSN: 0022-1287.

DOCUMENT TYPE: Article

FILE SEGMENT: BA

LANGUAGE: ENGLISH

AB *A. P. fluorescens* strain, isolated from a freshwater source on a plastic substratum, was grown in continuous culture in minimal medium. The adsorbable (adsorptive bubble separation) process foam-fractionated, wild-type cells from the fermenter during flow conditions. This selection pressure favored the enrichment of 2 major classes of mutant, both having cell surface characteristics fundamentally different from the wild-type. The wild-type produced very little extracellular polysaccharide. A mucoid mutant, found predominantly in the aqueous phase, produced an alginate exopolymer. The 2nd class of mutant was isolated from the walls of the fermenter and, like the wild-type, produced little exopolymer. This mutant, with crenated colony morphology, showed increased attachment to solid surfaces compared to the wild-type and mucoid cells when assayed for attachment to polystyrene surfaces for 2 h. Outer-membrane protein, lipopolysaccharides, and exopolysaccharides of the wild-type and both

mutants were analyzed. The results demonstrate the role of cell surface characteristics in the adaptability of the organism to micro-environments such as a solid/liquid or air/liquid interface or the aqueous phase.

L10 ANSWER 14 OF 38 BIOSIS COPYRIGHT (c) 2006 The Thomson Corporation on STN

ACCESSION NUMBER: 1982:178834 BIOSIS
DOCUMENT NUMBER: PREV198273038818; BA73:38818
TITLE: ISOLATION OF ALGINATE PRODUCING MUTANTS
OF PSEUDOMONAS-FLUORESCENS
PSEUDOMONAS-PUTIDA AND PSEUDOMONAS-MENDOCINA.
AUTHOR(S): GOVAN J R W [Reprint author]; FYFE J A M; JARMAN T R
CORPORATE SOURCE: DEP OF BACTERIOLOGY, UNIV OF EDINBURGH, MED SCHOOL, TEVIOT
PLACE, EDINBURGH EH8 9AG, UK
SOURCE: Journal of General Microbiology, (1981) Vol. 125, No. 1,
pp. 217-220.
CODEN: JGMIAN. ISSN: 0022-1287.
DOCUMENT TYPE: Article
FILE SEGMENT: BA
LANGUAGE: ENGLISH

AB Spontaneous alginate-producing (muc) variants were isolated from strains of *P. fluorescens*, *P. putida* and *P. mendocina* at a frequency of 1 in 108 by selecting for carbenicillin resistance. The IR spectrum of the bacterial exopolysaccharide was typical of an acetylated alginate similar to that previously described in *Azotobacter vinelandii* and in mucoid variants of *P. aeruginosa*. Mucoid variants were not isolated from *P. stutzeri*, *P. pseudoalcaligenes*, *P. testosteroni*, *P. diminuta*, *P. acidovorans*, *P. cepacia* or *P. maltophilia*.

L10 ANSWER 15 OF 38 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2004:101282 CAPLUS
DOCUMENT NUMBER: 140:162433
TITLE: Engineering *Pseudomonas fluorescens* variants for improved alginate production for use in cosmetics, pharmaceuticals and nutrients and animal feed
INVENTOR(S): Gimmestad, Martin; Sletta, Havard; Karunakaran, Karuna Ponniah; Bakkevig, Karianne; Ertesvag, Helga; Ellingsen, Trond; Skjak-Braek, Gudmund; Valla, Svein
PATENT ASSIGNEE(S): FMC Biopolymer AS, Norway
SOURCE: PCT Int. Appl., 76 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2004011628	A1	20040205	WO 2003-NO257	20030724
W:	AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW			
RW:	GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG			
CA 2493638	AA	20040205	CA 2003-2493638	20030724
AU 2003248516	A1	20040216	AU 2003-248516	20030724
EP 1543105	A1	20050622	EP 2003-771512	20030724

R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT,
IE, SI, LT, LV, FI, RO, MK, CY, AL, TR, BG, CZ, EE, HU, SK

CN 1685037	A	20051019	CN 2003-822409	20030724
JP 2005533515	T2	20051110	JP 2004-524404	20030724
NO 2005000480	A	20050408	NO 2005-480	20050127
US 2006063237	A1	20060323	US 2005-522510	20050917

PRIORITY APPLN. INFO.:	NO 2002-3581	A	20020726
	WO 2003-NO257	W	20030724

AB It is described biol. pure bacterial cultures of mutant strains of *Pseudomonas fluorescens*, which produces large amts. of alginate. The alginate may contain a certain determined content of mannuronate and guluronate residues, possible presence and determined level of acetyl groups in the alginate, and a desired mol. weight of the alginate. Also high yielding mutants with regulation of alginate production, is described. The invention further provides methods for producing new mutant strains of *Pseudomonas fluorescens* and variants thereof, and use of the resulting strains in improved alginate prodn for use in drugs, cosmetics, animal feed and nutrients.

REFERENCE COUNT: 13 THERE ARE 13 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L10 ANSWER 16 OF 38 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2003:817271 CAPLUS

DOCUMENT NUMBER: 140:25307

TITLE: Biofilm formation at the air-liquid interface by the *Pseudomonas fluorescens* SBW25 wrinkly spreader requires an acetylated form of cellulose

AUTHOR(S): Spiers, Andrew J.; Bohannon, John; Gehrig, Stefanie M.; Rainey, Paul B.

CORPORATE SOURCE: Department of Plant Sciences, University of Oxford, Oxford, OX1 3RB, UK

SOURCE: Molecular Microbiology (2003), 50(1), 15-27

CODEN: MOMIEE; ISSN: 0950-382X

PUBLISHER: Blackwell Publishing Ltd.

DOCUMENT TYPE: Journal

LANGUAGE: English

AB The wrinkly spreader (WS) genotype of *Pseudomonas fluorescens* SBW25 colonizes the air-liquid interface of spatially structured microcosms resulting in formation of a thick biofilm. Its ability to colonize this niche is largely due to overprodn. of a cellulosic polymer, the product of the wss operon. Chemical anal. of the biofilm matrix shows that the cellulosic polymer is partially acetylated cellulose, which is consistent with predictions of gene function based on in silico anal. of wss. Both polar and non-polar mutations in the sixth gene of the wss operon (wssF) or adjacent downstream genes (wssGHIJ) generated mutants that overproduce non-acetylated cellulose, thus implicating WssFGHIJ in acetylation of cellulose. WssGHI are homologs of AlgFIJ from *P. aeruginosa*, which together are necessary and sufficient to acetylate alginate polymer. WssF belongs to a newly established Pfam family and is predicted to provide acyl groups to WssGHI. The role of WssJ is unclear, but its similarity to MinD-like proteins suggests a role in polar localization of the acetylation complex. Fluorescent microscopy of Calcofluor-stained biofilms revealed a matrix structure composed of networks of cellulose fibers, sheets and clumped material. Quant. analyses of biofilm structure showed that acetylation of cellulose is important for effective colonization of the air-liquid interface: mutants identical to WS, but defective in enzymes required for acetylation produced biofilms with altered phys. properties. In addition, mutants producing non-acetylated cellulose were unable to spread rapidly across solid surfaces. Inclusion in these assays of a WS mutant with a defect in the GGDEF regulator (WspR) confirmed the requirement for this protein in expression of both acetylated

cellulose polymer and bacterial attachment. These results suggest a model in which WspR regulation of cellulose expression and attachment plays a role in the co-ordination of surface colonization.

REFERENCE COUNT: 52 THERE ARE 52 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L10 ANSWER 17 OF 38 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2003:466304 CAPLUS

DOCUMENT NUMBER: 139:335185

TITLE: The *Pseudomonas fluorescens* AlgG protein, but not its mannuronan C-5-epimerase activity, is needed for alginate polymer formation

AUTHOR(S): Gimmetstad, Martin; Sletta, Havard; Ertesvag, Helga; Bakkevig, Karianne; Jain, Sumita; Suh, Sang-jin; Skjok-Braek, Gudmund; Ellingsen, Trond E.; Ohman, Dennis E.; Valla, Svein

CORPORATE SOURCE: Department of Biotechnology, Norwegian University of Science and Technology, Trondheim, Norway

SOURCE: Journal of Bacteriology (2003), 185(12), 3515-3523
CODEN: JOBAA; ISSN: 0021-9193

PUBLISHER: American Society for Microbiology

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Bacterial alginates are produced as 1-4-linked β -D-mannuronan, followed by epimerization of some of the mannuronic acid residues to α -L-guluronic acid. Here we report the isolation of four different epimerization-defective point mutants of the periplasmic *Pseudomonas fluorescens* mannuronan C-5-epimerase AlgG. All mutations affected amino acids conserved among AlgG-epimerases and were clustered in a part of the enzyme also sharing some sequence similarity to a group of secreted epimerases previously reported in *Azotobacter vinelandii*. An algG-deletion mutant was constructed and found to produce predominantly a dimer containing a 4-deoxy-L-erythro-hex-4-enopyranosyluronate residue at the nonreducing end and a mannuronic acid residue at the reducing end. The production of this dimer is the result of the activity of an alginate lyase, AlgL, whose in vivo activity is much more limited in the presence of AlgG. A strain expressing both an epimerase-defective (point mutation) and a wild-type epimerase was constructed and shown to produce two types of alginate mols.: one class being pure mannuronan and the other having the wild-type content of guluronic acid residues. This formation of two distinct classes of polymers in a genetically pure cell line can be explained by assuming that AlgG is part of a periplasmic protein complex.

REFERENCE COUNT: 51 THERE ARE 51 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L10 ANSWER 18 OF 38 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2001:826434 CAPLUS

DOCUMENT NUMBER: 136:364494

TITLE: Characterization of algG encoding C5-epimerase in the alginate biosynthetic gene cluster of *Pseudomonas fluorescens*

AUTHOR(S): Morea, Antonella; Mathee, Kalai; Franklin, Michael J.; Giacomini, Alessio; O'Regan, Michael; Ohman, Dennis E.

CORPORATE SOURCE: Biotechnology Centre, CRIBI, University of Padova, Padua, 35121, Italy

SOURCE: Gene (2001), 278(1-2), 107-114
CODEN: GENED6; ISSN: 0378-1119

PUBLISHER: Elsevier Science B.V.

DOCUMENT TYPE: Journal

LANGUAGE: English

AB The organization of the alginate gene cluster in *Pseudomonas fluorescens* was characterized. A bank of

genomic DNA from *P. fluorescens* was mobilized to a strain of *Pseudomonas aeruginosa* with a transposon insertion (*algJ::Tn501*) in the alginate biosynthetic operon that rendered it non-mucoid. Phenotypic complementation in this heterologous host was observed, and a complementing clone containing 32 kb of *P. fluorescens* DNA was obtained. Southern hybridization studies showed that genes involved in alginate biosynthesis (e.g. *algD*, *algG*, and *algA*) were approx. in the same order and position as in *P. aeruginosa*. When the clone was mobilized to a *P. aeruginosa* *algG* mutant that produced alginate as polymannuronate due to its C5-epimerase defect, complementation was observed and the alginate from the recombinant strain contained l-guluronate as determined by proton NMR spectroscopy. A sequence anal. of the *P. fluorescens* DNA containing *algG* revealed sequences similar to *P. aeruginosa* *algG* that were also flanked by *algE*- and *algX*-like sequences. The predicted *AlgG* amino acid sequence of *P. fluorescens* was 67% identical (80% similar) to *P. aeruginosa* *AlgG* and 60% identical (76% similar) to *Azotobacter vinelandii* *AlgG*. As in *P. aeruginosa*, *AlgG* from *P. fluorescens* appeared to have a signal sequence that would localize it to the periplasm where *AlgG* presumably acts as a C5-epimerase at the polymer level. Non-polar *algG* knockout mutants of *P. fluorescens* were defective in alginate production, suggesting a potential role for this protein in polymer formation.

REFERENCE COUNT: 33 THERE ARE 33 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L10 ANSWER 19 OF 38 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1995:833235 CAPLUS

DOCUMENT NUMBER: 123:220822

TITLE: *Pseudomonas fluorescens*
mutant strains for the biocontrol of plant
pathogenic fungi

INVENTOR(S): Lam, Stephen; Torkewitz, Nancy

PATENT ASSIGNEE(S): Ciba-Geigy A.-G., Switz.

SOURCE: PCT Int. Appl., 38 pp.

CODEN: PIXXD2

DOCUMENT TYPE: Patent

LANGUAGE: English

FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9520040	A1	19950727	WO 1995-IB23	19950111
W: AM, AU, BB, BG, BR, BY, CA, CN, CZ, EE, FI, GE, HU, JP, KG, KP, KR, KZ, LK, LR, LT, LV, MD, MG, MN, MX, NO, NZ, PL, RO, RU, SI, SK, TJ, TT, UA, US, UZ, VN				
RW: KE, MW, SD, SZ, AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG				
US 5496547	A	19960305	US 1994-185623	19940124
AU 9512789	A1	19950808	AU 1995-12789	19950111
AU 694923	B2	19980806		
EP 743980	A1	19961127	EP 1995-903897	19950111
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LI, LU, MC, NL, PT, SE				
CN 1139453	A	19970101	CN 1995-191311	19950111
JP 09508269	T2	19970826	JP 1995-519443	19950111
RU 2154943	C2	20000827	RU 1996-117351	19950111
PRIORITY APPLN. INFO.:			US 1994-185623	A2 19940124
			WO 1995-IB23	W 19950111

AB Mutant strains of *Pseudomonas* were isolated by transposon insertion mutation and screening which have enhanced biocontrol properties, particularly against *Rhizoctonia solani*. A plasmid pCIB116 (containing a Tn5-promoterless lac transposable element) suitable for transposon mutagenesis was constructed and transferred to *Pseudomonas*

strain CGA 267356 to generate a collection of insertion mutants. About 10,000 mutants were tested for their ability to inhibit growth of *Neurospora* in vitro, and selected strains were further tested for their ability to control infestation by *R. solani* of cotton in greenhouse tests. Two insertion mutants provided better disease control against *R. solani* than did wild-type strains. Mutant strains CGA 319115 and CGA 32170 were applied as granules consisting of a finely divided carrier (e.g., vermiculite) and a polymer layer layer, wherein the polymer is (a) a film-forming, water-soluble, and essentially non-crosslinked polymer or (b) a film-forming, structurally crosslinked, water-swellaable polysaccharide (e.g., κ -carrageenan or sodium alginate).

L10 ANSWER 20 OF 38 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1992:406044 CAPLUS

DOCUMENT NUMBER: 117:6044

TITLE: Bioconversion of vanillin into vanillic acid by *Pseudomonas fluorescens* strain BTP9: cell reactors and mutants study

AUTHOR(S): Bare, G.; Gerard, J.; Jacques, P.; Delaunois, V.; Thonart, P.

CORPORATE SOURCE: Cent. Wallon Biol. Ind., Univ. Liege, Liege; 4000, Belg.

SOURCE: Applied Biochemistry and Biotechnology (1992), 34-35, 499-510

CODEN: ABIBDL; ISSN: 0273-2289

DOCUMENT TYPE: Journal

LANGUAGE: English

AB The ability of a fluorescent *Pseudomonas* to bioconvert vanillin, a phenolic compound, to vanillic acid was investigated. Free and immobilized cell reactors were tested. With free cells, the optimal yield reaches 98% after 6.5 h of bioconversion. With cells immobilized in alginate beads, transformation rate is only 47% after 13 h of conversion. Nevertheless, a continuous immobilized cell reactor was used for 76 h. With this, the optimal yield is >80%. The effects of residence time and cell concentration of the alginate beads in the reactor over the reactor's productivity also were studied. Catabolically blocked mutants for vanillic acid degradation were searched. To screen these mutants, a new and very sensitive method was developed. The results of mutant screenings are discussed.

L10 ANSWER 21 OF 38 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1983:536524 CAPLUS

DOCUMENT NUMBER: 99:136524

TITLE: Selection of attachment mutants during the continuous culture of *Pseudomonas fluorescens* and relationship between attachment ability and surface composition

AUTHOR(S): Pringle, J. Howard; Fletcher, Madilyn; Ellwood, D. C.

CORPORATE SOURCE: Dep. Environ. Sci., University of Warwick, Coventry, CV4 7AL, UK

SOURCE: Journal of General Microbiology (1983), 129(8), 2557-69

CODEN: JGMIAN; ISSN: 0022-1287

DOCUMENT TYPE: Journal

LANGUAGE: English

AB A strain of *P. fluorescens* that had been isolated from a freshwater source on a plastic substratum was grown in continuous culture in minimal medium. The adsorptive bubble separation process foam-fractionated wild-type cells from the fermentor during flow conditions. This selection pressure favored the enrichment of 2 major classes of mutants, both having cell surface characteristics fundamentally different from the wild-type. The wild-type produced very little extracellular polysaccharide, whereas a mucoid mutant, found predominantly in the aqueous phase, produced an

alginate exopolymer. The 2nd class of mutant was isolated from the walls of the fermentor and, like the wild-type, produced little exopolymer. This mutant, with crenated colony morphol., showed increased attachment to solid surfaces compared to the wild-type and mucoid cells when assayed for attachment to polystyrene surfaces for 2 h. Outer-membrane protein, lipopolysaccharides, and exopolysaccharides of the wild-type and both mutants were analyzed. The results demonstrate the role of cell surface characteristics in the adaptability of the organism to microenvironments such as a solid/liquid or air/liquid interface or the aqueous phase.

L10 ANSWER 22 OF 38 LIFESCI COPYRIGHT 2006 CSA on STN

ACCESSION NUMBER: 2003:107538 LIFESCI

TITLE: Biofilm formation at the air-liquid interface by the *Pseudomonas fluorescens* SBW25 wrinkly spreader requires an acetylated form of cellulose

AUTHOR: Spiers, A.J.; Bohannon, J.; Gehrig, S.M.; Rainey, P.B.

CORPORATE SOURCE: Department of Plant Sciences, University of Oxford, South Parks Road, Oxford OX1 3RB, UK.; E-mail: andrew.spiers@plants.ox.ac.uk

SOURCE: Molecular Microbiology [Mol. Microbiol.], (20031000) vol. 50, no. 1, pp. 15-27.
ISSN: 0950-382X.

DOCUMENT TYPE: Journal

FILE SEGMENT: G; J

LANGUAGE: English

SUMMARY LANGUAGE: English

AB The wrinkly spreader (WS) genotype of *Pseudomonas fluorescens* SBW25 colonizes the air-liquid interface of spatially structured microcosms resulting in formation of a thick biofilm. Its ability to colonize this niche is largely due to overproduction of a cellulosic polymer, the product of the wss operon. Chemical analysis of the biofilm matrix shows that the cellulosic polymer is partially acetylated cellulose, which is consistent with predictions of gene function based on in silico analysis of wss. Both polar and non-polar mutations in the sixth gene of the wss operon (wssF) or adjacent downstream genes (wssGHIJ) generated mutants that overproduce non-acetylated cellulose, thus implicating WssFGHIJ in acetylation of cellulose. WssGHI are homologues of AlgFIJ from *P. aeruginosa*, which together are necessary and sufficient to acetylate alginate polymer. WssF belongs to a newly established Pfam family and is predicted to provide acyl groups to WssGHI. The role of WssJ is unclear, but its similarity to MinD-like proteins suggests a role in polar localization of the acetylation complex. Fluorescent microscopy of Calcofluor-stained biofilms revealed a matrix structure composed of networks of cellulose fibres, sheets and clumped material. Quantitative analyses of biofilm structure showed that acetylation of cellulose is important for effective colonization of the air-liquid interface: mutants identical to WS, but defective in enzymes required for acetylation produced biofilms with altered physical properties. In addition, mutants producing non-acetylated cellulose were unable to spread rapidly across solid surfaces. Inclusion in these assays of a WS mutant with a defect in the GGDEF regulator (WspR) confirmed the requirement for this protein in expression of both acetylated cellulose polymer and bacterial attachment. These results suggest a model in which WspR regulation of cellulose expression and attachment plays a role in the co-ordination of surface colonization.

L10 ANSWER 23 OF 38 LIFESCI COPYRIGHT 2006 CSA on STN

ACCESSION NUMBER: 2003:73780 LIFESCI

TITLE: The *Pseudomonas fluorescens* AlgG Protein, but Not Its Mannuronan C-5- Epimerase Activity, Is Needed for Alginate Polymer Formation

AUTHOR: Gimmestad, M.; Sletta, H.; Ertesvaag, H.; Bakkevig, K.;

Jain, S.; Suh, S.; Skjåk-Braek, G.; Ellingsen, T.E.;
Ohman, D.E.; Valla, S.*
CORPORATE SOURCE: Department of Biotechnology, NTNU Norwegian University of
Science and Technology, N-7491 Trondheim, Norway; E-mail:
svein.valla@biotech.ntnu.no
SOURCE: Journal of Bacteriology [J. Bacteriol.], (20030600) vol.
185, no. 12, pp. 3515-3523.
ISSN: 0021-9193.
DOCUMENT TYPE: Journal
FILE SEGMENT: G; J
LANGUAGE: English
SUMMARY LANGUAGE: English

AB Bacterial alginates are produced as 1-4-linked beta
-D-mannuronan, followed by epimerization of some of the mannuronic acid
residues to [alpha] -L-guluronic acid. Here we report the isolation of
four different epimerization-defective point mutants of the
periplasmic *Pseudomonas fluorescens* mannuronan C-5-
epimerase AlgG. All mutations affected amino acids conserved among AlgG-
epimerases and were clustered in a part of the enzyme also sharing some
sequence similarity to a group of secreted epimerases previously reported
in *Azotobacter vinelandii*. An algG-deletion mutant was
constructed and found to produce predominantly a dimer containing a
4-deoxy-L-erythro-hex-4- enepyranosyluronate residue at the nonreducing
end and a mannuronic acid residue at the reducing end. The production of
this dimer is the result of the activity of an alginate lyase,
AlgL, whose in vivo activity is much more limited in the presence of AlgG.
A strain expressing both an epimerase-defective (point mutation) and a
wild-type epimerase was constructed and shown to produce two types of
alginate molecules: one class being pure mannuronan and the other
having the wild-type content of guluronic acid residues. This formation of
two distinct classes of polymers in a genetically pure cell line can be
explained by assuming that AlgG is part of a periplasmic protein complex.

L10 ANSWER 24 OF 38 LIFESCI COPYRIGHT 2006 CSA on STN

ACCESSION NUMBER: 2002:23850 LIFESCI
TITLE: Characterization of algG encoding C5-epimerase in the
alginate biosynthetic gene cluster of
Pseudomonas fluorescens
AUTHOR: Morea, A.; Mathee, K.; Franklin, M.J.; Giacomini, A.;
O'Regan, M.; Ohman, D.E.
CORPORATE SOURCE: CRIBI, Biotechnology Centre, University of Padova, Viale G.
Colombo 3, 35121 Padova Italy
SOURCE: Gene, (20011031) vol. 278, no. 1-2, pp. 107-114.
ISSN: 0378-1119.
DOCUMENT TYPE: Journal
FILE SEGMENT: G; J
LANGUAGE: English
SUMMARY LANGUAGE: English

AB The organization of the alginate gene cluster in
Pseudomonas fluorescens was characterized. A bank of
genomic DNA from *P. fluorescens* was mobilized to a strain of *Pseudomonas*
aeruginosa with a transposon insertion (algJ::Tn501) in the
alginate biosynthetic operon that rendered it non-mucoid.
Phenotypic complementation in this heterologous host was observed, and a
complementing clone containing 32 kb of *P. fluorescens* DNA was obtained.
Southern hybridization studies showed that genes involved in
alginate biosynthesis (e.g. algD, algG, and algA) were
approximately in the same order and position as in *P. aeruginosa*. When the
clone was mobilized to a *P. aeruginosa* algG mutant that produced
alginate as polymannuronate due to its C5-epimerase defect,
complementation was observed and the alginate from the
recombinant strain contained 1 -guluronate as determined by proton nuclear
magnetic resonance spectroscopy. A sequence analysis of the *P. fluorescens*
DNA containing algG revealed sequences similar to *P. aeruginosa* algG that

were also flanked by algE- and algX-like sequences. The predicted AlgG amino acid sequence of *P. fluorescens* was 67% identical (80% similar) to *P. aeruginosa* AlgG and 60% identical (76% similar) to *Azotobacter vinelandii* AlgG. As in *P. aeruginosa*, AlgG from *P. fluorescens* appeared to have a signal sequence that would localize it to the periplasm where AlgG presumably acts as a C5-epimerase at the polymer level. Non-polar algG knockout mutants of *P. fluorescens* were defective in alginate production, suggesting a potential role for this protein in polymer formation.

L10 ANSWER 25 OF 38 LIFESCI COPYRIGHT 2006 CSA on STN

ACCESSION NUMBER: 93:90362 LIFESCI

TITLE: Influence of *Pseudomonas fluorescens*
on hyphal growth and biocontrol activity of *Trichoderma harzianum* in the spermosphere and rhizosphere of pea.

AUTHOR: Dandurand, L.M.; Knudsen, G.R.

CORPORATE SOURCE: Plant Pathol. Div., Univ. Idaho, Moscow, ID 83843, USA

SOURCE: PHYTOPATHOLOGY., (1993) vol. 83, no. 3, pp. 265-270.

ISSN: 0031-949X.

DOCUMENT TYPE: Journal

FILE SEGMENT: A; K; J

LANGUAGE: English

SUMMARY LANGUAGE: English

AB *Trichoderma harzianum* isolate ThzID1 was grown in liquid culture, was formulated with alginate and polyethylene glycol 8000, and was milled into fine granules (average diameter 500 μ m). Granules contained chlamydospores, conidia, and hyphal fragments. Viability of the encapsulated fungus remained high for at least 6 mo when stored at 5 C (i.e., >90% of the granules produced hyphal growth when incubated on agar); viability was reduced significantly when granules were stored at 22 C. Application of the granular formulation of *T. harzianum* to pea seeds reduced root rot by *Aphanomyces euteiches* f. sp. pisi in growth-chamber experiments and also increased plant top weights compared to noncoated seeds. Seed treatment with slurries of *Pseudomonas fluorescens* strain 2-79RN sub(10), which produces a phenazine antibiotic, also reduced *Aphanomyces* root rot but to a lesser extent than did *T. harzianum* ThzID1. Disease suppression was not significantly different when seeds were treated with a combination of *T. harzianum* and 2-79RN sub(10) compared to treatment with *T. harzianum* alone. Root rot was not reduced by the mutant *P. fluorescens* strain 2-79-B46, which lacks phenazine. Treatment with *T. harzianum* plus 2-79-B46 resulted in the same level of disease control achieved by *T. harzianum* alone. These results suggest that the biocontrol mechanism of *P. fluorescens* 2-79RN sub(10) neither inhibited nor enhanced the biocontrol activity of *T. harzianum* ThzID1. In other experiments, density of *T. harzianum* hyphae originating from coated pea seeds in soil was not affected by the addition of 2-79RN sub(10), but when 2-79-B46 was added, density was greater after 5 days. The colony radius of *T. harzianum* was initially enhanced (at 3 days) by the addition of either strain, but the effect diminished by day 5. The same treatments were then applied to peas and to glass beads of equivalent size, and similar effects of the added bacterial strains were observed on both substrates, suggesting that the growth enhancement of *T. harzianum* in the presence of bacteria was not the direct result of stimulation of seed exudation by the bacteria. Our results provide a potentially improved formulation methodology for coating seeds with biocontrol organisms and methods for evaluating the compatibility of fungal and bacterial biocontrol agents applied to seeds.

L10 ANSWER 26 OF 38 LIFESCI COPYRIGHT 2006 CSA on STN

ACCESSION NUMBER: 93:7774 LIFESCI

TITLE: Bioconversion of vanillin into vanillic acid by
Pseudomonas fluorescens strain BTP9:
Cell reactors and mutants study.

PROCEEDINGS OF THE THIRTEENTH SYMPOSIUM ON BIOTECHNOLOGY

FOR FUELS AND CHEMICALS.

AUTHOR: Bare, G.; Gerard, J.; Jacques, P.; Delaunois, V.; Thonart, P.

CORPORATE SOURCE: Cent. Wallon Biol. Ind., Univ. Liege, F.S.A. Gx, Sart-Tilman, B40, 4000 Liege, Belgium

SOURCE: APPL. BIOCHEM. BIOTECHNOL., (1992) pp. 499-512.
Meeting Info.: 13. Symposium on Biotechnology for Fuels and Chemicals. (np).

DOCUMENT TYPE: Book

TREATMENT CODE: Conference

FILE SEGMENT: A; W

LANGUAGE: English

SUMMARY LANGUAGE: English

AB The ability of a fluorescent *Pseudomonas* to bioconvert vanillin, a phenolic compound, into vanillic acid was investigated. Free and immobilized cell reactors were tested. With free cells, the optimal yield reaches 98% after 6 h and 30 min of bioconversion. With cells immobilized in alginate beads, transformation rate is only 47% after 13 h of conversion. Nevertheless, a continuous immobilized cell reactor was used for 76 h. With this one, the optimal yield is higher than 80%. The influence of the residence time and cell concentration of the alginate beads in the reactor over the reactor's productivity has also been studied. Catabolically blocked mutants for vanillic acid degradation were searched.

L10 ANSWER 27 OF 38 LIFESCI COPYRIGHT 2006 CSA on STN

ACCESSION NUMBER: 93:5127 LIFESCI

TITLE: Influence of an antagonistic strain of *Pseudomonas fluorescens* on growth and ability of *Trichoderma harzianum* to colonize sclerotia of *Sclerotinia sclerotiorum* in soil.

AUTHOR: Bin, L.; Knudsen, G.R.; Eschen, D.J.

CORPORATE SOURCE: Plant Pathol. Div., Dep. Plant, Soil, and Entomol. Sci., Univ. Idaho, Moscow, ID 83843, USA

SOURCE: PHYTOPATHOLOGY., (1991) vol. 81, no. 9, pp. 994-1000.

DOCUMENT TYPE: Journal

FILE SEGMENT: J; A; W; K

LANGUAGE: English

SUMMARY LANGUAGE: English

AB *Pseudomonas fluorescens* strain 2-79RN sub(10) (nalidixic acid and rifampicin-resistant mutant of wild type strain 2-79) was used to study potential effects of bacterial antagonism in soil on growth and biocontrol efficacy of the biocontrol fungus *Trichoderma harzianum* isolate ThzID1, which was formulated into alginate pellets. In steamed soil (25 C, -100 or -500 kPa matric potential), strain 2-79RN sub(10) maintained its initial high populations (approximately 3×10^4 or 3×10^7 cfu/g of soil) over a 14-day period, and significantly reduced hyphal radius, hyphal density, and recoverable numbers of propagules of ThzID1. In raw soil under similar environmental conditions (22-25 C, -10 to -1,000 kPa), populations of 2-79RN sub(10) decreased by approximately four log sub(10) units over a 3-wk period, and did not affect the ability of *Trichoderma* spp. to colonize sclerotia of *Sclerotinia sclerotiorum*. Populations of 2-79RN sub(10) decreased gradually after 1-2 wk and did not reduce the ability of *Trichoderma* spp. to colonize sclerotia of *S. sclerotiorum*. Colonization of sclerotia by *Trichoderma* spp. after 9 wk was significantly higher in steamed soil (mean = 65%) than in raw soil (mean = 30%).

L10 ANSWER 28 OF 38 LIFESCI COPYRIGHT 2006 CSA on STN

ACCESSION NUMBER: 83:41158 LIFESCI

TITLE: Selection of attachment mutants during the continuous culture of *Pseudomonas fluorescens* and relationship between attachment ability and surface composition.

AUTHOR: Pringle, J.H.; Fletcher, M.; Ellwood, D.C.
CORPORATE SOURCE: Dep. Environ. Sci., Univ. Warwick, Coventry CV4 7AL, UK
SOURCE: J. GEN. MICROBIOL., (1983) vol. 129, no. 8, pp. 2557-2569.
DOCUMENT TYPE: Journal
FILE SEGMENT: J; M
LANGUAGE: English
SUMMARY LANGUAGE: English

AB A strain of *P. fluorescens* that had been isolated from a freshwater source on a plastic substratum was grown in continuous culture in minimal medium. The "adsubble" process (adsorptive bubble separation process) was found to foam-fractionate wild-type cells from the fermenter during flow conditions. This selection pressure favoured the enrichment of two major classes of mutant, both having cell surface characteristics fundamentally different from the wild-type. The wild-type produced very little extracellular polysaccharide, whereas a "mucoid" mutant, found predominantly in the aqueous-phase, produced an alginate exopolymer. Outer-membrane protein, lipopolysaccharides and exopolysaccharides of the wild-type and both mutants were analysed. The results demonstrate the role of cell surface characteristics in the adaptability of the organism to micro-environments such as a solid/liquid or air/liquid interface or the aqueous phase.

L10 ANSWER 29 OF 38 LIFESCI COPYRIGHT 2006 CSA on STN

ACCESSION NUMBER: 81:18399 LIFESCI

TITLE: Isolation of Alginate-Producing Mutants
of *Pseudomonas fluorescens*, *Pseudomonas*
putida and *Pseudomonas mendocina*.

AUTHOR: Govan, J.R.W.; Fyfe, J.A.M.; Jarman, T.R.

CORPORATE SOURCE: Dept. Bacteriol., Univ. Edinburgh, Med. Sch., Teviot Place,
Edinburgh EH8 9AG, UK

SOURCE: J. GEN. MICROBIOL., (1981) vol. 125, no. 1, pp. 217-220.

DOCUMENT TYPE: Journal

FILE SEGMENT: J

LANGUAGE: English

SUMMARY LANGUAGE: English

AB Spontaneous alginate-producing (muc) variants were isolated from strains of *P. fluorescens*, *P. putida* and *P. mendocina* at a frequency of 1 in 10 super(8) by selecting for carbenicillin resistance. The infrared spectrum of the bacterial exopolysaccharide was typical of an acetylated alginate similar to that previously described in *Azotobacter vinelandii*) and in mucoid variants of *P. aeruginosa*. Mucoid variants were not isolated from *P. stutzeri*, *P. pseudoalcaligenes*, *P. testosteroni*, *P. diminuta*, *P. acidovorans*, *P. cepacia* or *P. maltophilia*.

L10 ANSWER 30 OF 38 BIOTECHNO COPYRIGHT 2006 Elsevier Science B.V. on STN

ACCESSION NUMBER: 2003:37222783 BIOTECHNO

TITLE: Biofilm formation at the air-liquid interface by the
Pseudomonas fluorescens SBW25
wrinkly spreader requires an acetylated form of
cellulose

AUTHOR: Spiers A.J.; Bohannon J.; Gehrig S.M.; Rainey P.B.

CORPORATE SOURCE: A.J. Spiers, Department of Plant Sciences, University
of Oxford, South Parks Road, Oxford OX1 3RB, United
Kingdom.

E-mail: andrew.spiers@plants.ox.ac.uk

SOURCE: Molecular Microbiology, (2003), 50/1 (15-27), 52
reference(s)

CODEN: MOMIEE ISSN: 0950-382X

DOCUMENT TYPE: Journal; Article

COUNTRY: United Kingdom

LANGUAGE: English

SUMMARY LANGUAGE: English

AN 2003:37222783 BIOTECHNO

AB The wrinkly spreader (WS) genotype of *Pseudomonas*

fluorescens SBW25 colonizes the air-liquid interface of spatially structured microcosms resulting in formation of a thick biofilm. Its ability to colonize this niche is largely due to overproduction of a cellulosic polymer, the product of the wss operon. Chemical analysis of the biofilm matrix shows that the cellulosic polymer is partially acetylated cellulose, which is consistent with predictions of gene function based on in silico analysis of wss. Both polar and non-polar mutations in the sixth gene of the wss operon (wssF) or adjacent downstream genes (wssGHIJ) generated mutants that overproduce non-acetylated cellulose, thus implicating WssFGHIJ in acetylation of cellulose. WssGHI are homologues of AlgFIJ from *P. aeruginosa*, which together are necessary and sufficient to acetylate alginate polymer. WssF belongs to a newly established Pfam family and is predicted to provide acyl groups to WssGHI. The role of WssJ is unclear, but its similarity to MinD-like proteins suggests a role in polar localization of the acetylation complex. Fluorescent microscopy of Calcofluor-stained biofilms revealed a matrix structure composed of networks of cellulose fibres, sheets and clumped material. Quantitative analyses of biofilm structure showed that acetylation of cellulose is important for effective colonization of the air-liquid interface: mutants identical to WS, but defective in enzymes required for acetylation produced biofilms with altered physical properties. In addition, mutants producing non-acetylated cellulose were unable to spread rapidly across solid surfaces. Inclusion in these assays of a WS mutant with a defect in the GGDEF regulator (WspR) confirmed the requirement for this protein in expression of both acetylated cellulose polymer and bacterial attachment. These results suggest a model in which WspR regulation of cellulose expression and attachment plays a role in the co-ordination of surface colonization.

L10 ANSWER 31 OF 38 BIOTECHNO COPYRIGHT 2006 Elsevier Science B.V. on STN
 ACCESSION NUMBER: 2003:36676417 BIOTECHNO
 TITLE: The *Pseudomonas fluorescens* AlgG protein, but not its mannuronan C-5-epimerase activity, is needed for alginate polymer formation
 AUTHOR: Gimmetstad M.; Sletta H.; Ertesvag H.; Bakkevig K.; Jain S.; Suh S.-J.; Skjak-Braek G.; Ellingsen T.E.; Ohman D.E.; Valla S.
 CORPORATE SOURCE: S. Valla, Department of Biotechnology, NTNU Norwegian Univ. Sci./Technol., N-7491 Trondheim, Norway.
 E-mail: svein.valla@biotech.ntnu.no
 SOURCE: Journal of Bacteriology, (2003), 185/12 (3515-3523), 51 reference(s)
 CODEN: JOBAAY ISSN: 0021-9193
 DOCUMENT TYPE: Journal; Article
 COUNTRY: United States
 LANGUAGE: English
 SUMMARY LANGUAGE: English
 AN 2003:36676417 BIOTECHNO
 AB Bacterial alginates are produced as 1-4-linked β -D-mannuronan, followed by epimerization of some of the mannuronic acid residues to α -L-guluronic acid. Here we report the isolation of four different epimerization-defective point mutants of the periplasmic *Pseudomonas fluorescens* mannuronan C-5-epimerase AlgG. All mutations affected amino acids conserved among AlgG-epimerases and were clustered in a part of the enzyme also sharing some sequence similarity to a group of secreted epimerases previously reported in *Azotobacter vinelandii*. An algG-deletion mutant was constructed and found to produce predominantly a dimer containing a 4-deoxy-L-erythro-hex-4-enepyranosyluronate residue at the nonreducing end and a mannuronic acid residue at the reducing end. The production of this dimer is the result of the activity of an alginate lyase, AlgL, whose in vivo activity is much more limited in the presence of

AlgG. A strain expressing both an epimerase-defective (point mutation) and a wild-type epimerase was constructed and shown to produce two types of alginate molecules: one class being pure mannuronan and the other having the wild-type content of guluronic acid residues. This formation of two distinct classes of polymers in a genetically pure cell line can be explained by assuming that AlgG is part of a periplasmic protein complex.

L10 ANSWER 32 OF 38 BIOTECHNO COPYRIGHT 2006 Elsevier Science B.V. on STN
 ACCESSION NUMBER: 2001:33055784 BIOTECHNO
 TITLE: Characterization of algG encoding C5-epimerase in the alginate biosynthetic gene cluster of *Pseudomonas fluorescens*
 AUTHOR: Morea A.; Mathee K.; Franklin M.J.; Giacomini A.; O'Regan M.; Ohman D.E.
 CORPORATE SOURCE: D.E. Ohman, Department of Microbiology, M.C.V.C. Virginia Commonwealth Univ., 5-047 Sanger Hall, 1101 E. Marshall Street, Richmond, VA 23298-0678, United States.
 E-mail: deohman@hsc.vcu.edu
 SOURCE: Gene, (31 OCT 2001), 278/1-2 (107-114), 33 reference(s)
 CODEN: GENED6 ISSN: 0378-1119
 PUBLISHER ITEM IDENT.: S0378111901006850
 DOCUMENT TYPE: Journal; Article
 COUNTRY: Netherlands
 LANGUAGE: English
 SUMMARY LANGUAGE: English

AN 2001:33055784 BIOTECHNO
 AB The organization of the alginate gene cluster in *Pseudomonas fluorescens* was characterized. A bank of genomic DNA from *P. fluorescens* was mobilized to a strain of *Pseudomonas aeruginosa* with a transposon insertion (algJ::Tn501) in the alginate biosynthetic operon that rendered it non-mucoid. Phenotypic complementation in this heterologous host was observed, and a complementing clone containing 32 kb of *P. fluorescens* DNA was obtained. Southern hybridization studies showed that genes involved in alginate biosynthesis (e.g. algD, algG, and algA) were approximately in the same order and position as in *P. aeruginosa*. When the clone was mobilized to a *P. aeruginosa* algG mutant that produced alginate as polymannuronate due to its C5-epimerase defect, complementation was observed and the alginate from the recombinant strain contained L-guluronate as determined by proton nuclear magnetic resonance spectroscopy. A sequence analysis of the *P. fluorescens* DNA containing algG revealed sequences similar to *P. aeruginosa* algG that were also flanked by algE- and algX-like sequences. The predicted AlgG amino acid sequence of *P. fluorescens* was 67% identical (80% similar) to *P. aeruginosa* AlgG and 60% identical (76% similar) to *Azotobacter vinelandii* AlgG. As in *P. aeruginosa*, AlgG from *P. fluorescens* appeared to have a signal sequence that would localize it to the periplasm where AlgG presumably acts as a C5-epimerase at the polymer level. Non-polar algG knockout mutants of *P. fluorescens* were defective in alginate production, suggesting a potential role for this protein in polymer formation. .COPYRGHT. 2001 Elsevier Science B.V. All rights reserved.

L10 ANSWER 33 OF 38 EMBASE COPYRIGHT (c). 2006 Elsevier B.V. All rights reserved on STN
 ACCESSION NUMBER: 2005568101 EMBASE
 TITLE: Role of the *Pseudomonas fluorescens* alginate lyase (AlgL) in clearing the periplasm of alginates not exported to the extracellular environment.
 AUTHOR: Bakkevig K.; Sletta H.; Gimmestad M.; Aune R.; Ertesvag H.;

CORPORATE SOURCE: Degnes K.; Christensen B.E.; Ellingsen T.E.; Valla S.
S. Valla, Department of Biotechnology, Norwegian University
of Science and Technology (NTNU), N-7491 Trondheim, Norway.
svein.valla@biotech.ntnu.no
SOURCE: Journal of Bacteriology, (2005) Vol. 187, No. 24, pp.
8375-8384. .
Refs: 42
ISSN: 0021-9193 CODEN: JOBAAY
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
046 Environmental Health and Pollution Control
LANGUAGE: English
SUMMARY LANGUAGE: English
ENTRY DATE: Entered STN: 12 Jan 2006
Last Updated on STN: 12 Jan 2006

AB Alginate is an industrially widely used polysaccharide produced
by brown seaweeds and as an exopolysaccharide by bacteria belonging to the
genera *Pseudomonas* and *Azotobacter*. The polymer is composed of the two
sugar monomers mannuronic acid and guluronic acid (G), and in all these
bacteria the genes encoding 12 of the proteins essential for synthesis of
the polymer are clustered in the genome. Interestingly, 1 of the 12
proteins is an alginate lyase (AlgL), which is able to degrade
the polymer down to short oligouronides. The reason why this lyase is
associated with the biosynthetic complex is not clear, but in this paper
we show that the complete lack of AlgL activity in *Pseudomonas*
fluorescens in the presence of high levels of alginate
synthesis is toxic to the cells. This toxicity increased with the level
of alginate synthesis. Furthermore, alginate
synthesis became reduced in the absence of AlgL, and the polymers
contained much less G residues than in the wild-type polymer. To explain
these results and other data previously reported in the literature, we
propose that the main biological function of AlgL is to degrade
alginates that fail to become exported out of the cell and thereby
become stranded in the periplasmic space. At high levels of
alginate synthesis in the absence of AlgL, such stranded polymers
may accumulate in the periplasm to such an extent that the integrity of
the cell is lost, leading to the observed toxic effects. Copyright
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L10 ANSWER 34 OF 38 EMBASE COPYRIGHT (c) 2006 Elsevier B.V. All rights
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ACCESSION NUMBER: 2003405575 EMBASE
TITLE: Biofilm formation at the air-liquid interface by the
Pseudomonas fluorescens SBW25 wrinkly
spreader requires an acetylated form of cellulose.
AUTHOR: Spiers A.J.; Bohannon J.; Gehrig S.M.; Rainey P.B.
CORPORATE SOURCE: A.J. Spiers, Department of Plant Sciences, University of
Oxford, South Parks Road, Oxford OX1 3RB, United Kingdom.
andrew.spiers@plants.ox.ac.uk
SOURCE: Molecular Microbiology, (2003) Vol. 50, No. 1, pp. 15-27. .
Refs: 52
ISSN: 0950-382X CODEN: MOMIEE
COUNTRY: United Kingdom
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
LANGUAGE: English
SUMMARY LANGUAGE: English
ENTRY DATE: Entered STN: 23 Oct 2003
Last Updated on STN: 23 Oct 2003

AB The wrinkly spreader (WS) genotype of *Pseudomonas*
fluorescens SBW25 colonizes the air-liquid interface of spatially
structured microcosms resulting in formation of a thick biofilm. Its
ability to colonize this niche is largely due to overproduction of a

cellulosic polymer, the product of the wss operon. Chemical analysis of the biofilm matrix shows that the cellulosic polymer is partially acetylated cellulose, which is consistent with predictions of gene function based on in silico analysis of wss. Both polar and non-polar mutations in the sixth gene of the wss operon (wssF) or adjacent downstream genes (wssGHIJ) generated mutants that overproduce non-acetylated cellulose, thus implicating WssFGHIJ in acetylation of cellulose. WssGHI are homologues of AlgFIJ from *P. aeruginosa*, which together are necessary and sufficient to acetylate alginate polymer. WssF belongs to a newly established Pfam family and is predicted to provide acyl groups to WssGHI. The role of WssJ is unclear, but its similarity to MinD-like proteins suggests a role in polar localization of the acetylation complex. Fluorescent microscopy of Calcofluor-stained biofilms revealed a matrix structure composed of networks of cellulose fibres, sheets and clumped material. Quantitative analyses of biofilm structure showed that acetylation of cellulose is important for effective colonization of the air-liquid interface: mutants identical to WS, but defective in enzymes required for acetylation produced biofilms with altered physical properties. In addition, mutants producing non-acetylated cellulose were unable to spread rapidly across solid surfaces. Inclusion in these assays of a WS mutant with a defect in the GGDEF regulator (WspR) confirmed the requirement for this protein in expression of both acetylated cellulose polymer and bacterial attachment. These results suggest a model in which WspR regulation of cellulose expression and attachment plays a role in the co-ordination of surface colonization.

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ACCESSION NUMBER: 2003227419 EMBASE

TITLE: The *Pseudomonas fluorescens* AlgG protein, but not its mannuronan C-5-epimerase activity, is needed for alginate polymer formation.

AUTHOR: Gimmestad M.; Sletta H.; Ertesvag H.; Bakkevig K.; Jain S.; Suh S.-J.; Skjak-Braek G.; Ellingsen T.E.; Ohman D.E.; Valla S.

CORPORATE SOURCE: S. Valla, Department of Biotechnology, NTNU Norwegian Univ. Sci./Technol., N-7491 Trondheim, Norway.
svein.valla@biotech.ntnu.no

SOURCE: Journal of Bacteriology, (2003) Vol. 185, No. 12, pp. 3515-3523. .
Refs: 51

ISSN: 0021-9193 CODEN: JOBAAY

COUNTRY: United States

DOCUMENT TYPE: Journal; Article

FILE SEGMENT: 004 Microbiology
029 Clinical Biochemistry

LANGUAGE: English

SUMMARY LANGUAGE: English

ENTRY DATE: Entered STN: 26 Jun 2003

Last Updated on STN: 26 Jun 2003

AB Bacterial alginates are produced as 1-4-linked β -D-mannuronan, followed by epimerization of some of the mannuronic acid residues to α -L-guluronic acid. Here we report the isolation of four different epimerization-defective point mutants of the periplasmic *Pseudomonas fluorescens* mannuronan C-5-epimerase AlgG. All mutations affected amino acids conserved among AlgG-epimerases and were clustered in a part of the enzyme also sharing some sequence similarity to a group of secreted epimerases previously reported in *Azotobacter vinelandii*. An algG-deletion mutant was constructed and found to produce predominantly a dimer containing a 4-deoxy-L-erythro-hex-4-enepyranosyluronate residue at the nonreducing end and a mannuronic acid residue at the reducing end. The production of this dimer is the result of the activity of an alginate lyase, AlgL,

whose in vivo activity is much more limited in the presence of AlgG. A strain expressing both an epimerase-defective (point mutation) and a wild-type epimerase was constructed and shown to produce two types of alginate molecules: one class being pure mannuronan and the other having the wild-type content of guluronic acid residues. This formation of two distinct classes of polymers in a genetically pure cell line can be explained by assuming that AlgG is part of a periplasmic protein complex.

L10 ANSWER 36 OF 38 EMBASE COPYRIGHT (c) 2006 Elsevier B.V. All rights reserved on STN

ACCESSION NUMBER: 2001403406 EMBASE
TITLE: Characterization of algG encoding C5-epimerase in the alginate biosynthetic gene cluster of *Pseudomonas fluorescens*.
AUTHOR: Morea A.; Mathee K.; Franklin M.J.; Giacomini A.; O'Regan M.; Ohman D.E.
CORPORATE SOURCE: D.E. Ohman, Department of Microbiology, M.C.V.C. Virginia Commonwealth Univ., 5-047 Sanger Hall, 1101 E. Marshall Street, Richmond, VA 23298-0678, United States. deohman@hsc.vcu.edu
SOURCE: Gene, (31 Oct 2001) Vol. 278, No. 1-2, pp. 107-114. .
Refs: 33
ISSN: 0378-1119 CODEN: GENED6
PUBLISHER IDENT.: S 0378-1119(01)00685-0
COUNTRY: Netherlands
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
LANGUAGE: English
SUMMARY LANGUAGE: English
ENTRY DATE: Entered STN: 30 Nov 2001
Last Updated on STN: 30 Nov 2001

AB The organization of the alginate gene cluster in *Pseudomonas fluorescens* was characterized. A bank of genomic DNA from *P. fluorescens* was mobilized to a strain of *Pseudomonas aeruginosa* with a transposon insertion (algJ::Tn501) in the alginate biosynthetic operon that rendered it non-mucoid. Phenotypic complementation in this heterologous host was observed, and a complementing clone containing 32 kb of *P. fluorescens* DNA was obtained. Southern hybridization studies showed that genes involved in alginate biosynthesis (e.g. algD, algG, and algA) were approximately in the same order and position as in *P. aeruginosa*. When the clone was mobilized to a *P. aeruginosa* algG mutant that produced alginate as polymannuronate due to its C5-epimerase defect, complementation was observed and the alginate from the recombinant strain contained L-guluronate as determined by proton nuclear magnetic resonance spectroscopy. A sequence analysis of the *P. fluorescens* DNA containing algG revealed sequences similar to *P. aeruginosa* algG that were also flanked by algE- and algX-like sequences. The predicted AlgG amino acid sequence of *P. fluorescens* was 67% identical (80% similar) to *P. aeruginosa* AlgG and 60% identical (76% similar) to *Azotobacter vinelandii* AlgG. As in *P. aeruginosa*, AlgG from *P. fluorescens* appeared to have a signal sequence that would localize it to the periplasm where AlgG presumably acts as a C5-epimerase at the polymer level. Non-polar algG knockout mutants of *P. fluorescens* were defective in alginate production, suggesting a potential role for this protein in polymer formation. .COPYRGT. 2001 Elsevier Science B.V. All rights reserved.

L10 ANSWER 37 OF 38 EMBASE COPYRIGHT (c) 2006 Elsevier B.V. All rights reserved on STN

ACCESSION NUMBER: 83218444 EMBASE
DOCUMENT NUMBER: 1983218444
TITLE: Selection of attachment mutants during the continuous culture of *Pseudomonas*

fluorescens and relationship between attachment
ability and surface composition.
AUTHOR: Pringle J.H.; Fletcher M.; Ellwood D.C.
CORPORATE SOURCE: Dep. Environ. Sci., Univ. Warwick, Coventry CV4 7AL, United
Kingdom
SOURCE: Journal of General Microbiology, (1983) Vol. 129, No. 8,
pp. 2557-2469. .
CODEN: JGMIAN
COUNTRY: United Kingdom
DOCUMENT TYPE: Journal
FILE SEGMENT: 004 Microbiology
LANGUAGE: English
ENTRY DATE: Entered STN: 9 Dec 1991
Last Updated on STN: 9 Dec 1991

AB A strain of *Pseudomonas fluorescens* that had been
isolated from a freshwater source on a plastic substratum was grown in
continuous culture in minimal medium. The 'adsubble' process (adsorptive
bubble separation process) was found to foam-fractionate wild-type cells
from the fermenter during flow conditions. This selection pressure
favoured the enrichment of two major classes of mutant, both
having cell surface characteristics fundamentally different from the
wild-type. The wild-type produced very little extracellular
polysaccharide, whereas a 'mucoid' mutant, found predominantly
in the aqueous-phase, produced an alginate exopolymer. The
second class of mutant was isolated from the walls of the
fermenter and, like the wild-type, produced little exopolymer. This
mutant, with crenated colony morphology, showed increased
attachment of solid surfaces compared to the wild-type and mucoid cells
when assayed for attachment to polystyrene surfaces for 2 h.
Outer-membrane protein, lipopolysaccharides and exopolysaccharides of the
wild-type and both mutants were analysed. The results
demonstrate the role of cell surface characteristics in the adaptability
of the organism to micro-environments such as a solid/liquid or air/liquid
interface or the aqueous phase.

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ACCESSION NUMBER: 81225420 EMBASE
DOCUMENT NUMBER: 1981225420
TITLE: Isolation of alginate-producing mutants
of *Pseudomonas fluorescens*, *Pseudomonas*
putida and *Pseudomonas mendocina*.
AUTHOR: Govan J.R.W.; Fyfe J.A.M.; Jarman T.R.
CORPORATE SOURCE: Dept. Bacteriol., Univ. Edinburgh Med. Sch., Edinburgh EH8
9AG, United Kingdom
SOURCE: Journal of General Microbiology, (1981) Vol. 125, No. 1,
pp. 217-220. .
CODEN: JGMIAN
COUNTRY: United Kingdom
DOCUMENT TYPE: Journal
FILE SEGMENT: 004 Microbiology
LANGUAGE: English
ENTRY DATE: Entered STN: 9 Dec 1991
Last Updated on STN: 9 Dec 1991

DATA NOT AVAILABLE FOR THIS ACCESSION N